

Electronic Supplementary Information

A peptide with a cysteine terminus: Probe for label-free fluorescent detection of thrombin activity

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S1. Synthesis of dC₁₂-AgNCs

The synthesis of DNA-AgNCs was based on a previously reported method.^{S1,S2} AgNCs were synthesized by mixing AgNO₃ with a DNA (dC₁₂) solution and by adding NaBH₄ with continuous shaking. In brief, 150 μ l of the DNA (dC₁₂) template (200 μ M) was mixed with 2724 μ l of phosphate buffer (2.0 mM, pH 7.4). Freshly prepared AgNO₃ aqueous solution (90 μ l; 2.0 mM) was added to this solution. The resulting solution was vigorously shaken for 30 s. After 15 min in an ice bath, 36 μ l of freshly prepared NaBH₄ aqueous solution (5.0 mM) was added to the solution. The mixture was vigorously shaken for 1min. The solutions were kept in the dark at room temperature for 2 h and then incubated overnight at 4°C.

S2. Sequence-dependent responses of dC₁₂-AgNCs to peptides

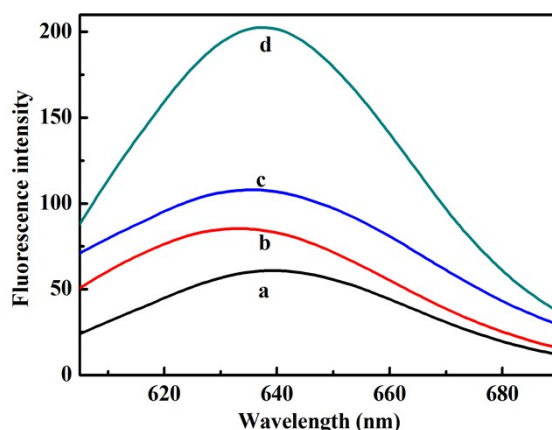


Fig. S1. Fluorescence responses of dC₁₂-AgNCs on different peptides (0.2 μ M). (a) dC₁₂-AgNCs. (b) dC₁₂-AgNCs + peptide 1. (c) dC₁₂-AgNCs + peptide 2. (d) dC₁₂-AgNCs + peptide 3.

S3. Optimizing the amount of GO.

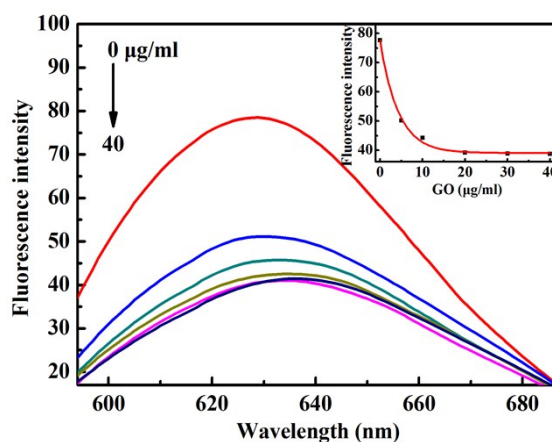


Fig. S2. Fluorescence responses of the dC₁₂-AgNCs-peptide 1 conjugate to different GO concentrations. 0.0, 10.0, 20.0, 30.0, and 40.0 μ g/mL. Inset: Changes in

fluorescence intensity at 635 nm as a function of GO concentration.

S4.Zeta potentials of GO

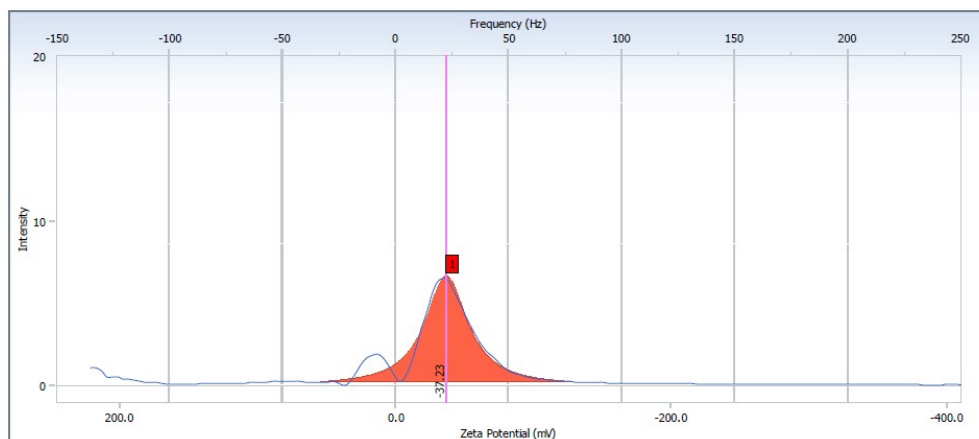


Fig. S3. Zeta potentials of 0.25 mg/mL GO in PBS (2.0 mM, pH= 7.4).

S5. Fluorescence responses of dC₁₂-AgNCs to peptides

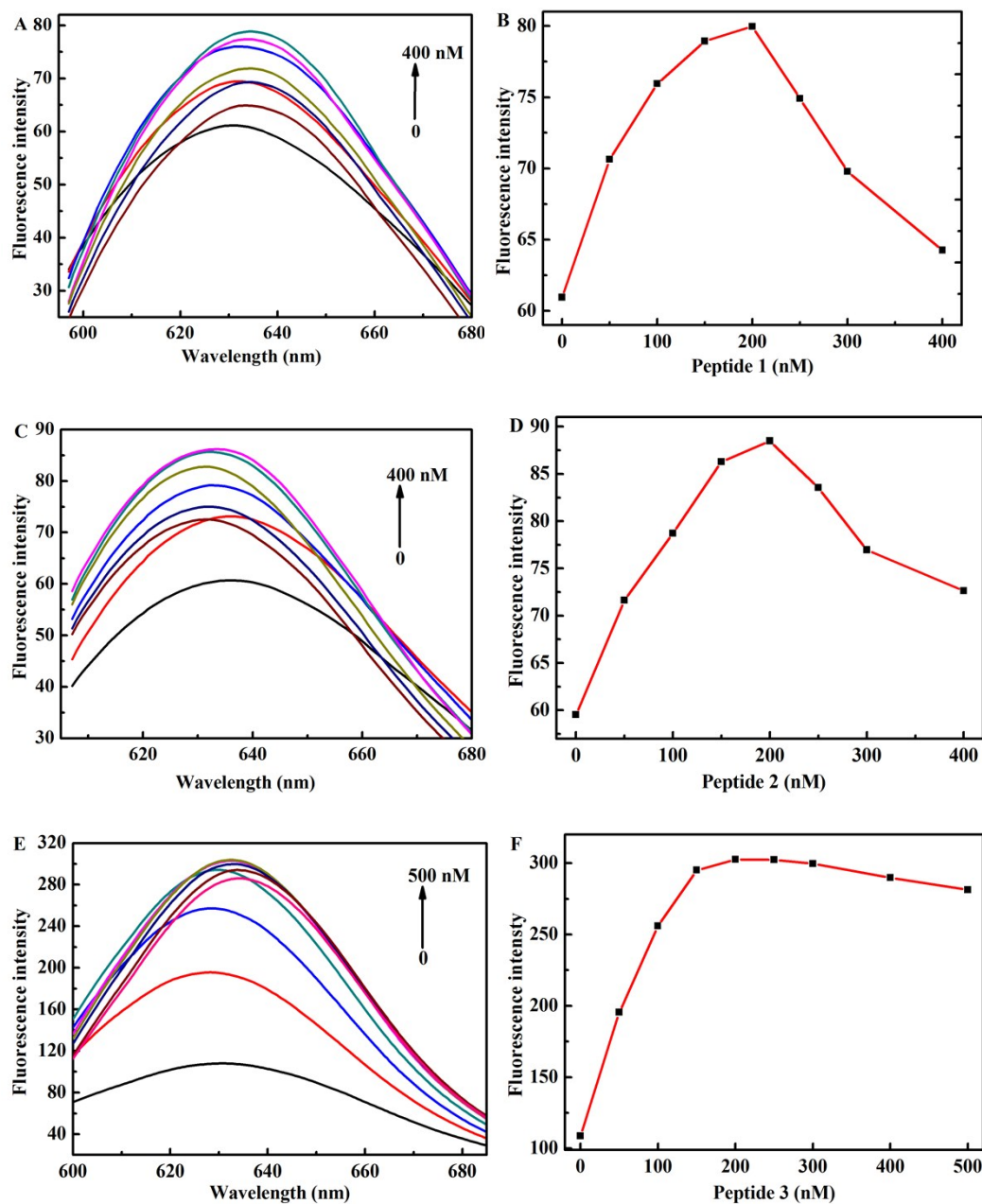


Fig. S4. Fluorescence responses of the dC₁₂-AgNCs to peptides. Peptide 1 (A), peptide 2 (C) and peptide 3 (E). Changes in fluorescence intensity at 635 nm versus the concentrations of peptides. Peptide 1 (B), peptide 2 (D) and peptide 3 (F).

S6. Fluorescence spectra of the ensemble of dC₁₂-AgNCs-peptide 2/GO incubated with thrombin

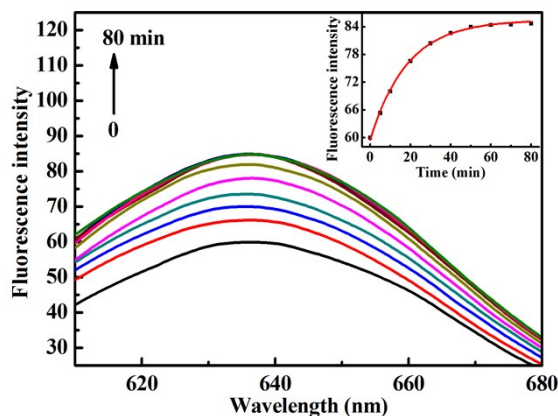


Fig. S5. Fluorescence spectra of the ensemble of dC₁₂-AgNCs-peptide 2/GO incubated with thrombin (0.1 μ M) at 37°C in different periods. Inset: Changes in fluorescence intensity at 635 nm as a function of incubation time.

S7. Fluorescence response of dC₁₂-AgNCs to 0.2 μ M peptides 1.

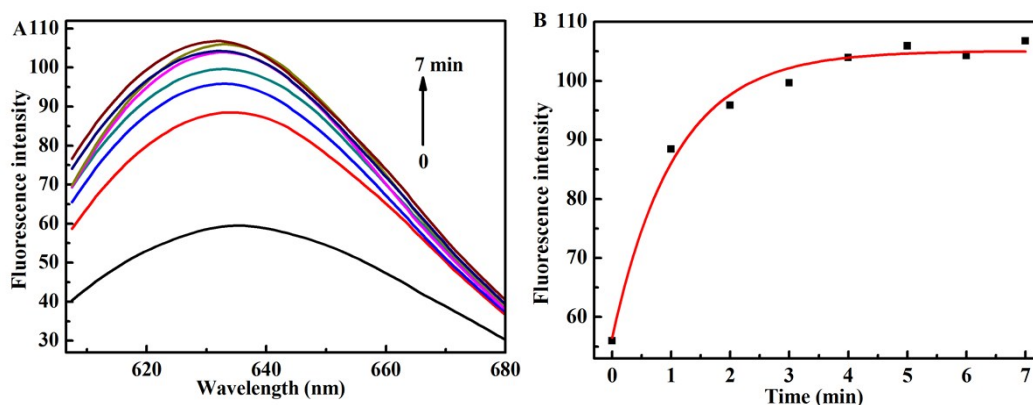


Fig. S6. (A) Fluorescence spectra of the dC₁₂-AgNCs reacted with peptide1 (0.2 μ M) at room temperature in different periods. (B) Changes in fluorescence intensity as a function of reacted time.

S8. Analytical performance of different platforms for the detection of thrombin.**Table S1 Analytical performance of different platforms for thrombin detection**

Method/System	Detection Limit (nM)	Detection time-interval/h	Ref.
Colorimetric, G-quadruplex DNAzyme	20	5	S3
Colorimetric, fibrinogen-gold nanoparticle	3.2×10^{-6}	8	S4
Fluorescence, Exo III-assisted analyte recycling	0.09	1-1.5	S5
Fluorescence, Aptamer-Functionalized Silver nanocluster DNA Probes	1	0.5	S6
Fluorescence, polymerization/nicking DNA machine	62.5	2	S7
Fluorescence, G-quadruplex-based DNAzyme	0.001	3	S8
ISFET, Nucleic acid modified GO matrices	25	1-2	S9
Absorbance, Aptamer-functionalized Au nanoparticles	2	3	S10
FRET, Upconverting Phosphors/Carbon Nanoparticles	0.18	1.25	S11
Bioluminescence, Sandwich-type immunoassay	10	1	S12
Electrochemiluminescence, Magnetic $\text{Fe}_3\text{O}_4@\text{CdSe}$ Composite Quantum Dot	1.2×10^{-2}	2	S13
Electrochemistry, Intercalation of MB in beacon aptamer	11	0.5	S14
Electrochemical impedance spectroscopy, Disposable screen- printed electrodes modified with functionalized graphene	10-50	1	S15
Fluorescence, GO/Aptamer/nucleic acid stabilized AgNCs	0.5	0.5-1	S16
Fluorescence, GO/peptide/nucleic acid stabilized AgNCs	1	1	Present study

S9. Reference SI

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